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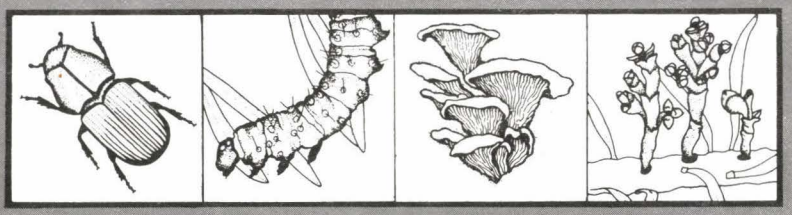

Report 86-11

3450
June 1986

ROOT DISEASES OF WESTERN WHITE PINE TRANSPLANTS AT THE USDA FOREST SERVICE NURSERY, COEUR D'ALENE, IDAHO

by

R. L. James, Plant Pathologist
and
C. J. Gilligan, Biological Technician

ABSTRACT

Mortality of western white pine transplants used to assess resistance to blister rust at the USDA Forest Service Nursery in Coeur d'Alene, Idaho was likely due to extensive root infection by Pythium spp. Severity of foliar symptom production was significantly correlated with extent of root system colonization by these fungi. Soil populations of Pythium were also related to disease occurrence which was most evident in poorly drained portions of beds. Although Fusarium oxysporum was frequently isolated from diseased transplants, its variable distribution in the soil and poor correlation with disease symptoms indicate that this fungus was probably not the major cause of the disorder.

INTRODUCTION

Containerized western white pine (Pinus monticola Dougl.) seedlings were transplanted several years ago at the USDA Forest Service Nursery, Coeur d'Alene, Idaho, as a progeny test to evaluate resistance to blister rust caused by Cronartium ribicola Fisch. Seedlings were transplanted into soil fumigated with methyl bromide/chloropicrin.

During the past 2 years, extensive areas of transplant mortality were noticed (figure 1). Most mortality seemed concentrated in groups, particularly in portions of beds that were poorly drained. There was also some scattered mortality, much of which occurred on individual transplants and was due to blister rust.

Previously, five diseased transplants from affected beds were collected and analyzed for occurrence of pathogens on their roots (James 1985b). Analyses were directed toward common soil-borne pathogens in the genus Fusarium. Only two of the five tested transplants had Fusarium on their roots. Other potential pathogens found were Cylindrocarpon and Verticillium. However, none of these fungi were consistently associated with the disease, at least not enough to be considered the major cause of mortality. Therefore, additional evaluations were necessary to elucidate etiology of the disorder.

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Due to financial constraints, only a limited number of photos were available.

Figure 1.--Transplanted western white pine seedlings exhibiting decline symptoms at the USDA Forest Service Nursery, Coeur d'Alene, Idaho. Affected transplants were usually concentrated in portions of beds which were poorly drained.

MATERIALS AND METHODS

One of the beds with several groups of diseased transplants (figure 1) was chosen for sampling. Fifteen transplants in various stages of decline were selected from within the sampled bed. Transplant foliar symptoms were rated (table 1) and transplant roots analyzed for presence of pathogens. Root systems were washed thoroughly under running tap water to remove soil. Root tips were aseptically cut from (1) the original containerized plug of roots and (2) the egressed lateral roots that arose from the plug. At least 15 root tips were sampled from each type of root system and incubated on selective media for Fusarium (Komada 1975) and Pythium (Hendrix and Kuhlman 1965). Fusarium plates were incubated under cool fluorescent light for 7 days at about 22° C and Pythium plates were incubated in the dark for 7 days at about 24° C. Percentage of root tips colonized with Fusarium and Pythium was determined.

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Table 1.--Numerical ratings of foliar conditions of western white pine transplants from the USDA Forest Service Nursery, Coeur d'Alene, Idaho.

Rating	Description
1.	Transplants tall; foliage dark green; appeared healthy.
2.	Transplants slightly dwarfed; foliage slightly chlorotic (or portions of foliage chlorotic).
3.	Transplants very dwarfed; foliage extensively chlorotic.
4.	Transplants dead; foliage red, indicating recent mortality.
5.	Transplants dead; foliage absent or gray, indicating older mortality.

In addition, soil samples were collected to determine populations of the two major groups of pathogens, Fusarium and Pythium. Samples were taken every 2 m along a transect starting at the west end of the sample bed. Twenty-five samples were taken in the middle of the bed and two samples were collected in the depression between this and adjacent beds. Each soil sample consisted of a cylindrical core of soil (2.5 cm diameter) placed directly in a paper bag. General condition (table 2) of the transplants near the soil sample site was assessed.

Table 2.--Numerical ratings of general transplant condition near soil sample sites at the USDA Forest Service Nursery, Coeur d'Alene, Idaho.

Rating	Description
1.	Most transplants dark green, tall, and appear healthy
2.	Most transplants slightly chlorotic and somewhat dwarfed
3.	Most transplants very chlorotic or necrotic and very dwarfed

Soil dilutions of 1:2 were made in 0.5 percent water agar (WA) and 1 ml dispensed onto plates of selective media for Pythium (Hendrix and Kuhlman 1965). Plates were incubated in the dark at 24°C for 72 hours after which Pythium colonies were counted. For determining Fusarium populations, soil dilutions of 1:400 were made in 0.1 percent WA and 1 ml dispensed onto plates of selective media for Fusarium (Komada 1975). Plates were incubated under cool, white fluorescent light continuously for 5 days at 24°C after which Fusarium colonies were counted. Pythium and Fusarium populations were calculated as colony-forming units (CFU) per gram of soil on the basis of each colony arising from one fungal propagule.

Fusarium isolates obtained from transplant roots and the soil samples were identified using the taxonomic guide of Nelson et al. (1983). No attempt was made to identify species of Pythium.

Linear regressions comparing numerical foliar condition ratings with extent of root colonization by Fusarium and Pythium were conducted. Regressions were also used to compare soil pathogen populations with general condition of nearby seedlings. All percentages underwent arc-sin transformations prior to analysis.

RESULTS

Table 3 summarizes extent of root colonization by Fusarium and Pythium on western white pine transplants displaying varying amounts of foliar necrosis. Generally, transplants with more severe foliar symptoms (higher numerical ratings) had their roots colonized to a greater extent by Pythium spp. Relationships between numerical foliar conditions rating and root colonization by Pythium were significant for both lateral ($P = 0.10$) and plug ($P = 0.005$) roots. However, significant relationships were not found for Fusarium colonization even though all sampled transplants had at least part of their root systems colonized by Fusarium spp.

Soil populations of Fusarium and Pythium within the sampled bed are summarized in table 4. Only two of the 25 samples yielded detectable populations of Fusarium; however, all but one produced colonies of Pythium. Comparisons between Pythium populations and general foliar condition of nearby transplants; i.e., severity of root disease symptoms, showed positive correlations which approached significance ($P = 0.25$). In other words, transplants with more severe disease symptoms were often in soil that had greater populations of Pythium.

Although all isolates of Fusarium obtained from infected transplant roots and soil samples were identified as Fusarium oxysporum Schlect., several different colony types were displayed by different isolates. For example, some exhibited deep purple staining and abundant flucose white aerial mycelium on potato dextrose agar, whereas others produced a slight purple pigment and light orange-colored mycelium. Tests to relate colony morphology to aggressiveness or pathogenic potential were not conducted.

DISCUSSION

On the basis of isolations from roots of transplants with disease symptoms and fungal populations in the soil, it seems likely that the major cause of western white pine transplant mortality was infection by Pythium spp. This conclusion was reinforced by the fact that much of the mortality occurred in poorly drained portions of beds, areas where damage to Pythium-like fungi are common (Vaartaja and Bumbieris 1964). Pythium-associated mortality has previously been described at the Coeur d'Alene Nursery on bareroot production stock (James 1982). Soil drenching with metalaxyl (Subdue®) effectively reduced damage and could probably be used to help prevent spread of the disease in transplant stock. However, much of the stock will soon be removed and beds will likely be fumigated to eliminate pathogens prior to replanting another crop.

Because transplants had been in beds for several years, it is possible that Pythium had reinvaded fumigated soil and increased in numbers over time, particularly in poorly drained portions of beds (Vaartaja 1967). Population counts of Pythium that we detected were not excessive compared to those reported for other conifer seedling nurseries (Marshall 1985; Vaartaja and Bumbieris 1964); however, they were apparently sufficient to cause extensive damage to transplant root systems.

Table 3.--Colonization of western white pine transplant roots by *Fusarium* spp. and *Pythium* spp. at the USDA Forest Service Nursery, Coeur d'Alene, Idaho.[1/]

Transplant condition rating[2/]	Fusarium			Pythium		
	Lateral[3/]	Plug[4/]	Total	Lateral[3/]	Plug[4/]	Total
1	0	20.0	9.1	29.4	6.7	18.8
1	9.8	14.9	12.0	21.4	6.2	14.9
1	7.1	0	3.6	30.8	27.9	29.3
Average (1)	5.6	11.6	8.2	27.2	13.6	21.0
2	5.4	0	2.8	21.2	21.6	21.4
2	0	16.7	8.4	21.6	20.0	20.8
2	2.4	2.3	2.3	27.5	15.5	21.2
2	5.3	2.3	3.7	28.6	43.2	36.0
Average (2)	3.3	5.3	4.3	24.7	25.1	24.8
3	43.3	0	21.7	33.3	16.1	24.2
3	3.2	16.7	9.7	20.0	26.7	23.3
3	0	3.3	1.7	15.6	13.3	14.5
3	9.8	22.9	15.8	50.0	52.6	51.3
Average (3)	14.1	10.7	12.2	29.7	27.2	28.3
4	6.7	0	3.3	26.7	25.8	26.2
4	9.5	0	4.8	70.7	42.5	56.8
4	4.6	0	2.4	54.3	52.5	53.5
Average (4)	6.9	0	3.5	50.6	40.3	45.5
5	15.8	2.8	9.5	39.5	61.3	49.3
Average (all transplants)	9.1	6.1	7.5	34.3	33.5	33.8
F value[5/]	--	--	0.004	4.301	11.614	9.149
Significance[5/]	—	—	NS	P=0.10	P=0.005	P=0.01

1/ Figures in table are percentages of sampled roots colonized.
2/ See table 1 for transplant condition ratings.
3/ Lateral roots are those that egressed from the original container plug.
4/ Roots within the original container plug.
5/ From linear regression analyses comparing root colonization by *Fusarium* and *Pythium* with numerical condition rating. Regressions were not conducted for lateral and plug colonization by *Fusarium* because of the nonsignificance (NS) of total root colonization.

Table 4.--Populations of *Fusarium* and *Pythium* in soil from western white pine transplant beds at the USDA Forest Service Nursery, Coeur d'Alene, Idaho.

General transplant condition[1/]	Population[2/] *Fusarium*	*Pythium*
1	0	3.3
1	0	4.7
1	0	4.0
1	0	2.0
1	0	10.7
1	0	0
1	0	8.0
1	0	10.0
1	0	4.7
1	0	2.7
1	0	2.7
1	0	10.0
Average (1)	0	5.2
2	133	9.3
2	0	4.7
2	0	8.7
2	0	2.7
2	667	4.0
2	0	2.7
2	0	6.7
2	0	14.0
2	0	5.3
2	0	8.7
Average (2)	80.0	6.7
3	0	5.3
3	0	10.0
3	0	9.3
Average (3)	0	8.2
Average all transplants	32.0	6.2
F value and significance[3/]	--	2.206 (P = 0.25)

1/ See table 2 for descriptions of general transplant conditions.

2/ Expressed as number of colony-forming units (CFU) per gram of soil.

3/ From linear regression analysis comparing *Pythium* soil populations with general condition of transplants near sample point. Analyses were not completed for *Fusarium* because of low counts.

Occurrence of F. oxysporum on roots of diseased transplants and populations detected in the soil were not consistent enough to conclude that this organism was the major cause of the disorder. This fungus is common in soils at the Coeur d'Alene Nursery (James and Gilligan 1985), but damage to bareroot seedlings is rare. It is possible that F. oxysporum colonized roots of transplants either after Pythium had infected or did so without eliciting disease symptoms, which commonly occurs (Bloomberg 1966). In any event, pathogenicity of F. oxysporum obtained from transplant stock was not tested and, therefore, its role in disease etiology remains questionable.

To reduce future problems in western white pine transplant beds at the Nursery, it is important that selected beds are well drained, that they are fumigated prior to transplanting, and that transplant stock is pathogen free. Since container stock is often used for transplants, care should be taken to select seedlings without root disease symptoms since diseased seedlings may die shortly after outplanting (James 1985a) or may serve as a source of infecting transplant soil with pathogens. If disease symptoms (other than blister rust) appear on transplants, spot soil drench treatments with metalaxyl should help restrict disease spread.

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